(iv) *Calculations*—(*a*) Calculate the micrograms of cefotetan per milligram of sample as follows:

$$\text{Micrograms of cefotetan per milligram} = \frac{A_u \times P_s \times 100}{A_s \times C_u \times (100 - m)}$$

where:

A_u=Area of the cefotetan peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_s=Area of the cefotetan peak in the chromatogram of the cefotetan working standard;

P_s=Cefotetan activity in the cefotetan working standard solution in micrograms per milliliter;

C_u=Milligrams of sample per milliliter of sample solution; and

m=Percent moisture content of the sample.

(*b*) Calculate the cefotetan content of the container as follows:

$$\text{Milligrams of cefotetan per container} = \frac{A_u \times P_s \times d}{A_s \times 1{,}000}$$

where:

A_u=Area of the cefotetan peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_s=Area of the cefotetan peak in the chromatogram of the cefotetan working standard;

P_s=Cefotetan activity in the cefotetan working standard solution in micrograms per milliliter; and

d=Dilution factor of the sample.

(2) *Sterility*. Proceed as directed in §436.20 of this chapter, using the method described in paragraph (e)(1) of that section.

(3) *Pyrogens.* Proceed as directed in §436.32(b) of this chapter, using a solution containing 50 milligrams of cefotetan per milliliter.

(4) *Moisture.* Proceed as directed in §436.201 of this chapter.

(5) *pH.* Proceed as directed in §436.202 of this chapter, using an aqueous solution containing 100 milligrams of cefotetan disodium per milliliter.

(6) *Identity*. The high-performance liquid chromatogram of the sample determined as directed in paragraph (b)(1) of this section, compares qualitatively to that of the cefotetan working standard.

[51 FR 20263, June 4, 1986, as amended at 52 FR 35912, Sept. 24, 1987; 55 FR 11583, Mar. 29, 1990]

§442.54 Cefpodoxime proxetil.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity*. Cefpodoxime proxetil is (±)-1-hydroxyethyl(+)-(6*R*,7*R*)-7-[2-(2-amino-4-thiazolyl)glyoxylamido]-3-(methoxymethyl)-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylate,7²-(*Z*)-(*O*-methyloxime), isopropyl carbonate (ester). It is so purified and dried that:

(i) Its potency is not less than 690 micrograms and not more than 804 micrograms of cefpodoxime activity per milligram, on an anhydrous basis.

(ii) The ratio of its R-epimer to total cefpodoxime is not less than 0.5 and not more than 0.6.

(iii) Its moisture content is not more than 3 percent.

(iv) It gives a positive identity test.

(2) *Labeling.* It shall be labeled in accordance with the requirements of §432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of §431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for cefpodoxime potency, isomer ratio, moisture, and identity.

(ii) Samples, if required by the Director, Center for Drug Evaluation and Research: 10 packages, each containing approximately 500 milligrams.

(b) *Tests and methods of assay*—(1) *Potency*. Proceed as directed in §436.216 of this chapter, using a suitable thermostatted column heating mechanism to maintain a column temperature of 40 °C, an ultraviolet detection system operating at a wavelength of 254 nanometers, a 15 centimeter X 4.6 millimeter (i.d.) column packed with microparticulate (5 micrometers in diameter) reversed phase packing material such as octadecyl silane bonded to silicas, a flow rate of 0.8 milliliter per minute, and a known injection volume of 2 microliters. The retention time for

the S-epimer is approximately 22 minutes and the retention time for R-epimer is approximately 28 minutes. The internal standard (propylparaben) has a retention time of 34 minutes. Mobile phase, dilution solvent, resolution solution, internal standard solution, working standard and sample solutions, system suitability requirements, and calculations are as follows:

(i) *Mobile phase.* The mobile phase consists of 420 milliliters of methanol, 580 milliliters of deionized water, and 230 milligrams of *L*-histidine hydrochloride. The pH is adjusted to 2.5±0.1 using 2*N* sulfuric acid. The mobile phase must be at room temperature for a correct pH measurement. The methanol concentration may be adjusted to achieve comparable retention times from column to column. Increasing methanol reduces retention times. Filter the mobile phase through a suitable filter capable of removing particulate matter 0.5 micron in diameter and degas it just before its introduction into the chromatograph.

(ii) *Dilution solvent.* Prepare a solvent for dilution by thoroughly mixing 495 milliliters of deionized water, 495 milliliters of acetonitrile, and 10 milliliters of acetic acid in an appropriate container.

(iii) *Resolution solution.* Prepare a 1 milligram per milliliter solution of any bulk containing ANTI–A in dilution solvent. Use this solution to determine the resolution between ANTI–A and the later-eluting drug epimer (R-epimer). Alternately, the resolution factor can be determined between the R and S isomers.

(iv) *Internal standard solution.* Prepare a solution of propylparaben in dilution solvent at a concentration of 10 milligrams per milliliter.

(v) *Preparation of working standard solutions.* Accurately weigh approximately 42 milligrams of the cefpodoxime proxetil working reference standard add 3 milliliters of internal standard solution and 25 milliliters of dilution solvent. The standard solution is stable for at least 48 hours. Refrigeration is not recommended.

(vi) *Sample solution.* Accurately weigh approximately 42 milligrams of the sample, add 3 milliliters of internal standard and 25 milliliters of dilution solvent. The sample solution is stable for at least 48 hours. Refrigeration is not recommended.

(vii) *System suitability requirements*—(A) *Asymmetry factor.*The asymmetry factor (A_s) is satisfactory if it is not less than 0.8 and not more than 1.1 for the R-epimer of cefpodoxime peak.

(B) *Efficiency of the column.* The absolute efficiency (h_r) is satisfactory if it is not more than 5 for the R-epimer peak.

(C) *Resolution factor.* The resolution factor (R) between the peak for ANTI–A and the peak for the R-epimer is satisfactory if it is not less than 1.3. Alternately, the resolution factor (R) between the peak for the R-epimer and the peak for the S-epimer of cefpodoxime is not less than 11.

(D) *Coefficient of variation (Relative standard deviation).* The coefficient of variation ($S_R$in percent of 5 replicate injections) is satisfactory if it is not more than 2 percent.

(E) *Capacity factor (k').* The capacity factor (k') for the R-epimer of cefpodoxime is satisfactory if it is not less than 10.4 and not more than 15.6.

(F) If the system suitability parameters in this paragraph (b)(1)(iv) have been met, then proceed as described in § 436.216(b) of this chapter.

(viii) *Calculations.* Calculate the micrograms of cefpodoxime proxetil per milligram of sample on an anhydrous basis as follows:

$$\text{Micrograms of cefpodoxime proxetil per milligram} = \frac{R_u \times P_s \times 100}{R_s \times C_u \times (100 - m)}$$

where:

R_u = Ratio of cefpodoxime proxetil peaks area (sum of both epimers) to the internal standard peak response in the sample solution;

R_s = Ratio of cefpodoxime proxetil peaks

area (sum of both epimers) to the internal standard peak response in the working standard solution;

P_s = Cefpodoxime proxetil activity of the working standard solution in micrograms per milliliter;

C_u = Milligrams of sample per milliliter of sample solution; and

m = Percent moisture content of the sample.

(2) *Isomer ratio.* Using the procedure described in paragraph (b)(1) of this section, calculate the ratio of the R-epimer (Ab) to the sum of the S-epimer and R-epimer (Aa and Ab), by the equation

$$\text{Isomer Ratio} = Ab/(Aa + Ab)$$

where:

Aa = Area of the early eluting S-epimer peak; and

Ab = Area of the late eluting R-epimer peak.

(3) *Moisture.* Proceed as directed in § 436.201 of this chapter, except use 30 milliliters of solvent C instead of 20 milliliters of solvent A.

(4) *Identity.* Proceed as directed in § 436.211 of this chapter, using the mineral oil mull prepared as described in paragraph (b)(2) of that section.

[60 FR 58231, Nov. 27, 1995]

§ 442.55 Ceftriaxone sodium.

(a) *Requirements for certification*—(1) *Standards of identity, strength, quality, and purity.* Ceftriaxone sodium is the 5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid, 7-[[(2-amino-4-thiazolyl)(methoxyimino)acetyl]amino]-8-oxo-3-[[(1,2,5,6-tetrahydro-2-methyl-5,6-dioxo-1,2,4-triazin-3-yl)thio]methyl]-,disodium salt, [6*R*-[6*alpha*, 7*beta*(*Z*)]]-. It is so purified and dried that:

(i) Its ceftriaxone potency is not less than 795 micrograms of ceftriaxone per milligram on an anhydrous free acid basis.

(ii) Its moisture content is not less than 8 percent and not more than 11 percent.

(iii) The pH of an aqueous solution containing the equivalent of 100.0 milligrams per milliliter is not less than 6.0 and not more than 8.0.

(iv) It is crystalline.

(v) It gives a positive identity test for ceftriaxone.

(2) *Labeling.* It shall be labeled in accordance with the requirements of § 432.5 of this chapter.

(3) *Requests for certification; samples.* In addition to complying with the requirements of § 431.1 of this chapter, each such request shall contain:

(i) Results of tests and assays on the batch for ceftriaxone potency, moisture, pH, crystallinity, and identity.

(ii) Samples, if required by the Director, Center for Drug Evaluation and Research: 10 packages, each containing approximately 500 milligrams.

(b) *Tests and methods of assay*—(1) *Ceftriaxone potency.* Proceed as directed in § 442.55a(b)(1) of this chapter, except prepare the sample solution and calculate the micrograms of ceftriaxone free acid per milligram as follows:

(i) *Preparation of sample solution.* Dissolve an accurately weighed portion of the sample with sufficient water to obtain a concentration of 180 micrograms of ceftriaxone activity per milliliter. Prepare the sample solution just prior to its introduction into the chromatograph.

(ii) *Calculation.* Calculate the micrograms of ceftriaxone anhydrous free acid per milligram as follows:

$$\text{Micrograms of ceftriaxone anhydrous free acid per milligram} = \frac{A_u \times P_s}{A_s \times C_u}$$

where:

A_u=Area of the ceftriaxone peak in the chromatogam of the sample (at a retention time equal to that observed for the standard);

A_s=Area of the ceftriaxone peak in the chromatogram of the ceftriaxone working standard;

P_s=Ceftriaxone activity in the ceftriaxone working standard solution in micrograms of anhydrous free acid per milliliter; and

C_u=Milligrams of sample per milliliter of sample solution.

(2) *Moisture.* Proceed as directed in § 436.201 of this chapter.

(3) *pH.* Proceed as directed in § 436.202 of this chapter, using an aqueous solution containing 100 milligrams per milliliter.

(4) *Crystallinity.* Proceed as directed in § 436.203(a) of this chapter.